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Gelation Characteristics of Tropical Surimi under Water Bath and Ohmic Heating

Panchaporn Tadpitchayangkoon[1], Jae W Park[2], Jirawat Yongsawatdigul[3]

[1]Department of Agro-Industry, Faculty of Agriculture,

Ubon Ratchathani University, Ubon Ratchathani, 34190, Thailand

[2]Oregon State University Seafood Laboratory,

2001 Marine Dr. # 253, Astoria, OR 97103, USA

[3]School of Food Technology, Institute of Agricultural Technology, Suranaree University of Technology, Nakhon Ratchasima, 30000, Thailand

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Corresponding author: Dr. Jirawat Yongsawatdigul

School of Food Technology, Institute of Agricultural Technology, Suranaree University of Technology, Nakhon Ratchasima, 30000, Thailand

Tel: 66-44-224359 Fax: 66-44-224150

Email: jirawat@sut.ac.th

ABSTRACT

Gelation characteristics of tropical surimi, namely threadfin bream (TB), bigeye snapper (BS), goatfish (GF) and lizardfish (LF) prepared in the absence and presence of 10 g kg^{-1} egg white proteins were evaluated using either ohmic (OH) or water bath (WB) heating. LF and GF surimi exhibited higher endogenous proteolytic activity than BS and TB. Ohmic heating markedly minimized proteolysis of LF and GF surimi as evidenced by a reduction of trichloroacetic acid (TCA)-soluble oligopeptide content of gels and more retention of myosin heavy chain (MHC). Ohmic heating increased breaking force and deformation of TB and BS surimi by 1.3 and 1.6 times, respectively, as compared to water bath heating. However, TB surimi gels heated by a higher applied voltage gradient of 16.7 V cm^{-1} exhibited lower breaking force than those heated at 6.7 V cm^{-1}. Gels heated ohmically contained lower total sulfhydryl concentration, indicating the greater extent of disulfide bond formation as compared to gels heated in a 90 °C water bath. The rapid heating method with shorter heating time could improve water holding capacity and preserve color of tropical surimi gels when compared to water bath heating.

Keywords: Ohmic heating, water bath heating, tropical surimi, gelation

1. Introduction

World production of tropical surimi has continually increased in the past decade, with an estimated 330 Gg in 2010. Important tropical fish used for surimi production includes threadfin bream (TB, *Nemipterus spp*.), bigeye snapper (BS, *Priacnathus spp*.), goatfish (GF, *Upeneus spp*.) and lizardfish (LF, *Saurida spp*.). According to a standard surimi gel testing method, surimi is chopped with salt and stuffed into a 3 cm-diameter casing and heated to 84-90 °C in a water bath for 30 min (National Fisheries Institute, 1991). Heating of surimi paste in 3-cm diameter casing requires 20-30 min to reach 90 °C in the center depending on the number of sausages and volume of heated water (Park & Lin, 2005). Although water bath heating is typically used for surimi gel evaluation, it does not accurately reflect gel formation of surimi during fast cooking for crabstick production. In commercial crabstick production, surimi paste is extruded as a thin sheet (1.2-2.2 mm) and heated directly by steam, gas, or electrical resistance. Heating rate in such a production line is about 100-120 °C min^{-1} (Park, 2005), which is markedly higher than the heating rate applied to water bath heating for surimi gel quality testing. Heating rate is known to have a great effect on gelation of muscle proteins (Barbut & Mittal, 1990; Arntfield & Murray, 1992; Cofrades, Carballo, & JimCnez-Colmenero, 1997; Sun & Arntfield, 2011). A method to evaluate gel quality and provide information relevant to gel formation at the production line is therefore necessary for better quality control.

Ohmic heating is a method that can achieve a fast heating rate. Heat is internally generated when alternating current is passed through an electrically conducting material, creating uniform temperature distribution (de Alwis & Fryer, 1990; Pataro, Donsì, & Ferrari, 2011). Ohmic cooking of bologna at varied heating rates of 3.9-10.3 °C min^{-1} resulted in comparable quality to the conventional heating method except for softer texture (Piette et al., 2004). An electric field in ohmic heating also enhanced inactivation of lipoxygenase and

polyphenoloxidase (Castro, Macedo, Teixeira, & Vicente, 2004). Only surimi from Alaska pollock and Pacific whiting, which are cold and temperate species, have been studied under ohmic heating thus far (Yongsawatdigul and Park 1996; Pongviratchai & Park, 2007). Gelation induced by ohmic heating greatly depends on intrinsic properties of individual surimi, particularly endogenous proteolytic activity. Intrinsic properties of tropical surimi are also known to vary with species and are different from those of cold/temperate species (Benjakul, Viessaguan, Thongkaew & Tanaka, 2003). Despite of the increasing volume of tropical surimi used in surimi seafood industry worldwide, its gelation under higher heating rates relevant to the real crabstick production line is unknown.

Egg white is a typical protein ingredient used in surimi seafood products including crabstick. Egg white proteins function as both a proteinase inhibitor and gel enhancer. It has been demonstrated that egg white proteins improve textural properties of surimi from various species due to its proteinase inhibitory activity (Piyachomkwan & Penner, 1995; Yongsawatdigul & Piyadhammaviboon, 2004; Benjakul, Visessanguan, Tueksuban, & Tanaka, 2004). In addition, egg white proteins positively contributed to surimi gel texture because of its own gel-forming ability (Li, Lin, & Kim, 2007; Hunt, Park, & Handa, 2009). However, the positive effect of egg white proteins on textural properties of surimi was entirely based on studies conducted using slow heating water bath. Therefore, the effect of egg white proteins on gelation of surimi under rapid heating should be investigated. Our objectives were first to measure the effect of water bath and ohmic heating on gelation of tropical surimi and further to elucidate physico-chemical changes of tropical surimi with addition of egg white proteins under both heating regimes.

2. Materials and methods

2.1 Tropical surimi samples

99 Frozen tropical surimi, namely threadfin beam (*Nemipterus spp.*), bigeye snapper
100 (*Priacanthus spp.)*, goatfish (*Upeneus spp.*), and lizardfish (*Saurida spp.*), were obtained from
101 Andaman Surimi Industry (Samutsakorn, Thailand). Surimi was cut into 500-g blocks,
102 vacuum-packed, and kept at -30 ^{o}C until used. All surimi samples contained 60 g kg^{-1} sucrose
103 and 3 g kg^{-1} sodium tripolyphosphate as cryoprotectants without addition of egg white proteins.
104 Spray-dried egg white (P-110) manufactured by Henningsen Foods (Ohama, Nebr., USA) was
105 used.

106 *2.2 Gel preparation*

107 Frozen surimi blocks were thawed at room temperature for about 2 h and cut into
108 approximately 5-cm cubes. The initial temperature of surimi was about 2-3 ^{o}C. Surimi cubes
109 were chopped without temperature control in a Stephan vacuum cutter (UM5, Stephan
110 Machinery Co., Columbus, OH, USA) at low speed for 1 min. Surimi pastes were prepared
111 either in the absence or presence of 10 g kg^{-1} egg white proteins, respectively. Dry ingredients
112 (either 20 g kg^{-1} salt or 20 g kg^{-1} salt+10 g kg^{-1} egg white proteins of total weight) were added
113 and chopping at low speed was continued for 1 min. Ice was added to adjust moisture
114 concentration to 780 g kg^{-1} and chopping continued an additional of 1 min on low speed (1,800
115 rpm). Subsequently, samples were chopped at high speed (3,600 rpm) for 3 min under vacuum
116 to a final temperature of 22-25 ^{o}C (Esturk, Park, & Thawornchinsombut, 2004). Final moisture
117 concentration of paste was measured using the standard oven method (AOAC, 1999). The paste
118 was vacuum-packed in a plastic bag to eliminate air bubbles and stuffed into cellulose casing (3
119 cm diameter) and nylon tubes (3 cm diameter) for water bath and ohmic heating, respectively.
120 The interior wall of the nylon tubes was sprayed with PAM cooking spray (Boyle-Midway Inc.,
121 NY, USA) before stuffing. Temperature of all pastes before heating was approximately 25 ^{o}C.

122 The samples were heated at 90 ^{o}C for 30 min for water bath heating. For ohmic heating,
123 the ohmic apparatus was described in details by Pongviratchai and Park (2007). An ohmic

heating apparatus was developed from two titanium electrodes at each end of nylon tube. The apparatus was connected to a voltage transducer, model VT8-007D and a current transducer, model CT8-015DY101 (Ohio Semitronics Inc., Hilliard, OH, USA). Temperature was minored using a T-type thermocouple and was controlled through a temperature controller (Model CNi3254-C24, Omega Engineering Inc., Stamford, CT, USA). The sample length exposed to ohmic heating was approximately 15 ± 0.5 cm. The titanium electrode with a diameter of 3.0 cm was inserted into each end of the suimi tube. The minimum pressure (276 kPa) was applied to assure sufficient contact between the electrode and surimi sample. The samples were heated to 90 ^{o}C at a frequency of 10 kHz at voltage levels of 100 and 250 V, corresponding to a voltage gradient of 6.7 (OH 6.7) and 16.7 (OH 16.7) V cm^{-1}, respectively. Heating time of the formers was 180.43±4.62 s., while of the latters was 40.06±1.44 s. A holding time at 90 ^{o}C for 60.06±1.04 s was maintained after set temperature at the center of sample was attained. Upon heating, gels were wrapped in a plastic film, cooled in iced water for 20 min and refrigerated overnight. All gel samples were analyzed within 48 h.

2.3 Trichloroacetic acid (TCA)-soluble oligopeptide content

TCA-soluble oligopeptide concentrations were measured according to the method described by Yongsawatdigul and Piyadhmmaviboon (2004). In this study, 65 °C was selected to evaluate proteolysis since this temperature has been reported to be the optimum for proteolytic activity of tropical fish muscle protein (Benjakul, Visessanguan, Ishizaki, & Tanaka, 2001; Yongsawatdigul, & Piyadhammaviboon, 2004; Benjakul, Visessanguan, & Tueksuban, 2003; Rawdkuen & Benjakul, 2008). To investigate the inhibitory effect of egg white proteins, paste samples with and without egg white proteins (3 g) were incubated at 65 ^{o}C for 1 h. The reaction was stopped by adding 27 ml of 50 g l^{-1} cold TCA solution. The mixtures were then homogenized and centrifuged at 10,000 × g for 15 min at 4 ^{o}C. The sample blank

148 was kept in ice and treated in the same manner as the samples. The supernatants were collected
149 and used for TCA-soluble oligopeptide concentration using Lowry's assay (Lowry,
150 Rosebrough, Farr, & Randall, 1951) with tyrosine as a standard. The samples were solubilized
151 in 50 g l^{-1} hot sodium dodecylsulfate (SDS) solution (1:9) and determined for total soluble
152 protein by Lowry's assay using bovine serum albumin (BSA) as a standard. Autolytic activity
153 of each species was expressed as nmol tyrosine mg $protein^{-1}$ h^{-1}.

154 TCA-soluble oligopeptide concentrations of gel samples were also determined. Three
155 grams of gel were added to 27 ml of 50 g l^{-1} cold TCA solution. The mixture was then
156 homogenized, centrifuged, and analyzed for oligopeptide concentration as described above.
157 TCA-soluble oligopeptide concentration was expressed as nmol tyrosine mg $protein^{-1}$

158 *2.4 SDS-PAGE pattern*

159 Protein patterns of the paste incubated at 65 ^{o}C for 1 h and cooked gels were determined
160 using SDS-PAGE (Laemmli, 1970). Stacking and separating gels were made of 40 and 100 g l^{-1}
161 polyacrylamide, respectively. Samples were solubilized in 50 g l^{-1} SDS solution. Protein
162 sample (20 μg) was loaded onto each lane on polyacrylamide gel. Gels were run at a constant
163 voltage setting at 200 V. Gels were stained with 1.25 g l^{-1} Coomassie Brilliant Blue R-250 and
164 destained in a solution containing 250 ml l^{-1} ethanol and 100 ml l^{-1} acetic acid.

165 *2.5 Texture measurement*

166 The chilled gels were left at room temperature for 2 h before texture and color
167 measurements. Textural properties of gels were measured using a Texture Analyzer (TA-XT
168 plus, Texture Technologies Corp. Scarsdale, NY, USA) equipped with a 5-mm diameter
169 spherical plunger probe. Gel samples were cut into 3 cm long pieces. Penetration test was

performed at a probe speed of 1 mm s^{-1}. Breaking force (g) and deformation (mm) were recorded. Ten measurements were made for each treatment and subjected to statistical analysis.

2.6 Total sulfhydryl (SH) concentration

Total SH concentrations of paste and cooked gels were determined according to Monahan, German, and Kinsella (1995). Samples were homogenized in a solubilizing buffer (0.2 mol l^{-1} Tris-HCl, 20 g l^{-1} SDS, 0.01 mol l^{-1} EDTA, 8 mol l^{-1} urea, pH 7.0). The homogenates were heated at 100 ^{o}C for 60 min and centrifuged at 10,000 × g for 15 min. Aliquot of the supernatant (1 ml) was added 0.01 ml Ellman's reagent (0.01 mol l^{-1} 5,5'-dinitrobis [2-nitrobenzoic acid]). The mixtures were incubated at 40 ^{o}C for 25 min. The absorbance measured at 412 nm was used to calculate total SH concentration using the extinction coefficient of 13600 M^{-1} cm^{-1}. Protein concentration was determined using Lowry's assay with BSA as a standard.

2.7 Water holding capacity (WHC)

WHC of all gels was evaluated according to Kang, Hunt, and Park (2008). Cooked gel cut into small pieces and 0.4±0.05 g was placed in a micro-centrifuge tube (Eppendrof, Hamburg, Germany) with a nylon screen and 0.45-μm pore size. Samples were then centrifuged at 5000 × *g* for 10 min at room temperature. The released water was weighted. Moisture content of cooked gel was measured using the standard oven method (AOAC, 1999). Water holding capacity was calculated as:

$$WHC\ (\%) = [(A-B)/C]*100$$

A = Weight of total water in surimi gel (g)

B = Weight of water released (g)

C = Weight of surimi gel (g)

193 *2.8 Color measurement*

194 Color values (L*, a*, b*) of all gel samples were measured using a colorimeter (Minolta
195 USA, Ramsey, NJ, USA). Whiteness of gel was calculated using the equation
196 L* - 3b* (Hunt, Park, & Handa 2009).

197 *2.9 Statistical analyses*

198 Two different lots of surimi were used. The experiment was analyzed as a split plot.
199 Four types of fish species (TB, BS, GF and LF) and two levels of egg white proteins (0 and 10
200 g kg^{-1}) were assigned as a main plot factor and 3 heating conditions (WB, OH 6.7 and OH 16.7)
201 as a split plot factor. In each treatment, at least 8 gel specimens were measured to obtain
202 average values of breaking force and deformation, 5 specimens for color, and 4 specimens for
203 others. Degree of variation and significance of difference were analyzed using analysis of
204 variance (ANOVA) with General Linear Models (SPSS for window, version 10.01 SPSS Inc,
205 Chicago, IL, USA). Duncan's multiple range test (DMRT) was used to determine differences
206 between means at $P < 0.05$.

207

208 **3. Results and discussion**

209 *3.1 Proteolysis*

210 TCA-soluble oligopeptide content reflects the extent of proteolytic degradation
211 resulting from endogenous proteinases of surimi. Endogenous proteolytic activity varies with
212 species (Table 1). Raw surimi of LF and GF exhibited the highest TCA-soluble oligopeptide
213 content ($P<0.05$), implying that they are proteinase-laden surimi. Myofibrillar-associated serine
214 proteinase was assumed to be present in LF surimi (Yongsawatdigul & Piyadhammaviboon,
215 2004). Proteinases from GF muscle were classified as cysteine and serine proteinases
216 (Yarnpakdee, Benjakul, Visessanguan, & Kijroongrjana, 2009). LF underwent severe myosin

heavy chain (MHC) degradation at 65 ^{o}C (Fig. 1a). It should be noted that the extent of actin degradation was less than MHC in LF surimi, indicating that MHC is a more preferred substrate. When 10 g kg^{-1} egg white proteins was incorporated into surimi pastes, TCA-soluble oligopeptide content of LF and GF pastes decreased ($P<0.05$, Table 1). This was most likely due to inhibitory activity towards endogenous proteinases of LF and GF surimi. Addition of 10 g kg^{-1} egg white proteins greatly protected MHC degradation of LF surimi (Fig. 1a). Inhibition of endogenous proteinase activity by egg white protein was less pronounced in TB and BS surimi pastes due to their low activity (Table 1).

As TB and BS surimi gels contained less proteolytic activity, addition of egg white proteins or fast ohmic heating did not affect TCA-soluble oligopeptide contents (Table 1) and MHC degradation (data not shown) of both species. In contrast, a fast heating rate significantly reduced proteolysis of LF and GF surimi gels. During water bath heating, endogenous proteinases of LF and GF surimi exhibiting optimum activity at 65 ^{o}C would actively hydrolyze muscle proteins during the slow thermal gradient before thermal inactivation of the proteinase occurred. In contrast, when rapid heating was applied, these enzymes were rapidly inactivated, rendering the limited proteolysis and low TCA-soluble oligopeptides. Retention of MHC of ohmically-cooked gels of all species was greater than that of the water bath-heated counterparts, especially in GF and LF gels (Fig. 1a,b).

Based on TCA-soluble oligopeptide contents, addition of egg white proteins along with the use of ohmic heating reduced proteolysis up to 58% in LF surimi, while applying either one approach reduced proteolytic activity by up to 27%. Retention of MHC of LZ samples was greater when egg white was added in conjunction with ohmic heating (Fig. 1a). Such a synergistic effect was not noticed in GF surimi gel. Proteolysis of GF surimi was inhibited only about 14 and 25% by addition of egg white proteins and ohmic heating, respectively. The combined effect of egg white proteins and ohmic heating only reduced proteolysis of GF up to

242 27%. MHC retention was not increased in the combined treatment as compared to samples
243 heated ohmically alone (Fig. 1b). A rapid heating rate seems to be a more effective means in
244 controlling endogenous proteinase activity of GF than addition of 10 g kg^{-1} egg white protein.
245 It could be speculated that egg white proteins (10 g kg^{-1}) did not effectively inhibit all major
246 proteinases involved in proteolysis of GF surimi.

247 The applied voltage gradients of 6.7 and 16.7 V cm^{-1} did not affect the extent of
248 proteolysis of all surimi gels studied ($P>0.05$, Table 1). This was likely because heating time of
249 approximately 180 and 40 s of voltage gradients of 6.7 and 16.7 V cm^{-1}, respectively, was
250 equally sufficient to inactivate endogenous proteinases of the studied tropical species.

251 *3.2 Textural properties*

252 In the absence of egg white proteins, TB surimi exhibited the highest breaking force
253 followed by BS, LF and GF, regardless of heating method applied ($P<0.05$, Fig. 2a). TB also
254 exhibited higher gel deformation than others while lower deformation was observed in GF and
255 LF gels ($P<0.05$). Gel-forming ability appeared to inversely correlate with the degree of
256 proteolysis (Table 1). Ohmic heating with a voltage gradient of 6.7 and 16.7 V cm^{-1} resulted in
257 gels with higher breaking force and deformation than those heated in a water bath for all
258 species tested ($P<0.05$). Breaking force and deformation of TB and BS surimi gels increased by
259 about 1.3-1.6 times when voltage gradient of 6.7 V cm^{-1} was applied. More striking
260 improvement was observed in GF gels heated ohmically with approximately 3.7- and 2.6-fold
261 increase in breaking force and deformation, respectively, as compared to those heated in a
262 water bath. LF surimi exhibited the lowest gel-forming ability when heated in a 90 °C-water
263 bath, corresponding to extensive loss of MHC induced by endogenous proteinases (Table 1 and
264 Fig. 1a). Ohmic heating was able to improve breaking force of LF gels by about 2 folds, while
265 deformation was comparable to those heated in a water bath. These results suggested that

ohmic heating significantly improved textural properties of tropical surimi gel without egg white proteins by minimizing the degree of proteolysis during heating.

The extent of textural improvement appeared to be greater in surimi associated with high endogenous proteolytic activity (GF, LF) by 2.6-3.7 times than in surimi with relatively low proteinase activity (TB, BS) by 1.3-1.6 times. Rapid heating inactivates proteinases fast, leading to more retention of intact myofibrillar proteins, particularly myosin, for gel network formation. Gel strength of surimi made from walleye pollock, white croaker, threadfin bream and sardine also increased after ohmic heating (Shiba, 1992; Shiba & Numakura, 1992). Extremely good gels were observed from Pacific whiting surimi after heated ohmically with an applied voltage gradient of 13.3 V cm^{-1} (Yongsawatdigul, Park, Kolbe, AbuDagga, & Morrissey, 1995). In general, the different heating rate in ohmic heating (30 and 135 $^{o}C\ min^{-1}$ for the applied voltage gradient of 6.7 and 16.7 V cm^{-1}, respectively) did not affect breaking force and deformation of tropical surimi except for TB whose breaking force appeared to reduce at high voltage gradient of 16.7 V cm^{-1}. As TB surimi exhibited the least proteolytic activity (Table 1), rapid thermal inactivation of endogenous proteinase is not the only factor governing gelation of TB surimi. Gel network also depends on alignment of unfolded proteins, which is a kinetically controlled process. A total heating time of approximately 40 s in the applied voltage gradient of 16.7 V cm^{-1} might have limited the development of gel network, rendering lower gel strength. Gel properties of turkey and Alaska pollock surimi heated from 10 to 70 ^{o}C exhibited less fracture stress when heated by microwave at 98 $^{o}C\ min^{-1}$ than at 20 $^{o}C\ min^{-1}$ (Riemann, Lanier, & Swartzel, 2004). The faster heating rates would allow less time at temperatures above the denaturation temperature of the protein for aggregation (Foegeding, Allen, & Dayton, 1986). This would explain why weaker gels were obtained at high heating rate of ohmic heating for TB surimi. It is, therefore, reasonable to state that extremely high

heating rate is not always beneficial for gelation of surimi with less endogenous proteolytic activity.

Addition of 10 g kg^{-1} egg white proteins increased breaking force and deformation of all gels heated in a water bath, particularly LF and GF surimi gels whose textural properties increased about 3 folds compared to those without addition of egg white proteins (Fig. 2a, b). This was mainly attributed to proteinase inhibitory activity and gel-forming ability of egg white proteins. Addition of egg white proteins at the level as low as 10 g kg^{-1} has also been reported to improve textural properties of Alaska pollock and Pacific whiting surimi (Hunt, Park, & Handa, 2009). Addition of 10 g kg^{-1} egg white proteins to LF surimi and pre-incubated at 25 ^{o}C prior to heating at 90 ^{o}C resulted in an increase in breaking force (Yongsawatdigul & Piyadhammaviboon, 2004). In addition, addition of other protein additives including chicken plasma proteins and fish sarcoplasmic proteins effectively minimized proteolysis and improved textural properties of BF and LF (Rawdkuen, Benjakul, Visessanguan, & Lanier, 2005; Piyadhammaviboon, & Yongsawatdigul, 2009). Ohmic heating also improved textural properties of TB, BS, and LF gels with addition of egg white proteins. However, breaking force and deformation values of GF surimi with addition of egg white proteins decreased when heated ohmically and were lower than those heated in a water bath ($P<0.05$). In addition, GF surimi with egg white proteins heated with higher heating rates of ohmic showed lower textural properties than those heated at slower heating rates ($P<0.05$). It was speculated that interactions between myofibrillar proteins of GF surimi and egg white proteins for proper gel network formation were not favorable under rapid heating regimes. Heat-induced aggregation of egg white proteins entirely depends on heating time and temperature (Naofumi, Shimizu, & Doi, 1988; Hsieh, Regenstein, & Handa, 1993). Therefore, interaction between fish myofibrillar proteins and other protein additives used in surimi seafood products in fast heating process can also be a critical parameter governing textural properties of the finished product.

For TB and BS surimi, addition of egg white proteins under ohmic heating did not increase textural properties further when compared to ohmic heating without egg white proteins. In a rapid heating process, addition of egg white proteins to these surimi samples is, therefore, not necessary for gel improvement. But egg white powder can enhance textural properties of surimi when subjected to a slow heating process. This is because proteinase inhibition is more critical for a slow heating regime. It is interesting to note that synergistic effect of egg white proteins and ohmic heating in inhibiting endogenous proteolytic activity of LF surimi was obvious (Table 1, Fig. 1a) and an effect on textural improvement was also pronounced (Fig. 2). Based on our study, a rapid heating process significantly improved textural properties of proteinase-laden surimi gel like LF or GF due to rapid thermal inactivation of endogenous proteinases. For surimi with less proteinase problem (TB surimi and other surimi with egg white proteins), rapid heating is still more preferable to slow heating, but sufficient heating time should be allowed for properly ordered aggregation which is vital to obtain high gel strength. Heating time of 3 min with applied voltage gradient of 6.7 V cm^{-1} resulted in superior texture quality compared to 40 s heating time of 16.7 V cm^{-1} for TB surimi with and without egg white proteins and GF surimi with addition of egg white proteins (Fig. 2).

Our results demonstrated that conventional surimi gel testing method using a 90 °C water bath, which has been widely used by industry and academia, obviously underestimates the quality of surimi, especially when such surimi is intended to be used for rapidly heated crabstick or fried surimi seafood. Water bath heating allows proteinases to actively degrade myofibrillar proteins, resulting in poor gel quality. The underestimation of gel quality is more pronounced in proteinase-laden surimi. Since ohmic heating provides the similar heating rate to the crabstick production at 110-120 $^{o}C\ min^{-1}$, the use of ohmic heating apparatus is highly recommended for accurate assessment of surimi gel quality.

3.3 Total SH concentration

Total SH content of TB, BS and GF gels without addition of egg white proteins decreased upon water bath heating (Fig. 3). Oxidation of sulfhydryl groups to form disulfide linkages occurs upon thermal gelation, resulting in a decrease of total SH content. Partial unfolding of actomyosin induced covalent cross-linking of reactive SH groups at 25 ^{o}C (Yongsawatdigul, Worratao, & Park, 2002) while major conformation changes of TB was found at greater than 35 ^{o}C and inter- and/or intra-molecular disulfide bonds were formed at > 50 ^{o}C (Yongsawatdigul & Park, 2003). Formation of disulfide bonds in natural actomyosin of tilapia was enhanced at > 75 ^{o}C (Ko, Yu, & Hsu, 2007). Total SH content of LF gel was not significantly decreased upon water bath heating. Severe proteolysis of LF muscle proteins, especially MHC, induced by slow heating in a 90 °C water bath (Table 1, Fig. 1a) resulted in oligopeptides, limiting gel network formation and disulfide bond formation.

Total SH content of surimi samples without egg white proteins decreased when subjected to ohmic heating as compared to water bath heating (Fig. 3, $P<0.05$). Ohmic heating obviously induced disulfide formation of tropical surimi samples. An increase in disulfide bonds of LF and GF surimi heated ohmically was explained by effective reduction of proteolysis under ohmic heating, leading to more available intact myofibrillar proteins for formation of disulfide bonds and other interactions important for gel network stabilization. For TB and BS, rapid heating might have induced conformational changes of myofibrillar proteins differently from water bath heating. To our knowledge, denaturation and aggregation pattern of myofibrillar proteins under a high heating rate of 135 ^{o}C min^{-1} equivalent to ohmic heating applied in this study has not been well elucidated. Such information would provide insight on how myofibrillar proteins aggregate to form networks under a rapid heating process.

Total SH content of all surimi samples with added egg white proteins decreased regardless of heating regimes applied. Addition of egg white proteins promoted disulfide formation by increasing protein-protein interactions between myofibrillar proteins and egg

white proteins as well as among egg white proteins themselves. Similar to samples without egg white proteins, total SH content of ohmically-cooked gels with egg white proteins reduced to a greater extent than those of water bath-cooked gels. These results revealed that rapid heating induced more protein unfolding, exposing more reactive SH which subsequently formed disulfide bonds during ohmic heating. These results imply that disulfide bonds favorably formed at fast heating could in part explain improvement of textural properties of ohmically-cooked surimi gels apart from rapid proteolytic inactivation.

3.4 Water holding capacity (WHC)

WHC of gels varied with surimi species and treatments ($P<0.05$, Fig. 4). TB surimi gel cooked ohmically with and without egg white proteins exhibited the highest WHC ($P<0.05$). Gels cooked by ohmic heating showed higher WHC than water bath- cooked gels ($P<0.05$). WHC correlated well with breaking force and deformation of surimi gels (Fig. 2). Our findings are in agreement with Shirsat, Brunton, and Lyng (2004) who reported that WHC of steam-cooked meat emulsion batter were significantly lower than those of samples cooked ohmically at 3, 5 or 7 V cm^{-1}. An increase in WHC was also found in a large diameter comminuted meat product cooked by a rapid heating method of radio frequency (Zang, Lyng, & Brunton, 2004).

Addition of egg white proteins also increased water holding properties of tropical surimi cooked in a water bath, especially LF and GF surimi. However, it did not increase WHC of gels heated ohmically. These results confirmed that gel network of the mixed myofibrillar and egg white proteins might not be fully developed to entrap water molecules under a rapid heating regime. Therefore, optimum heating rate to induce unfolding of egg white proteins would be required for 3-dimensional network formation of surimi–egg white proteins mixture under ohmic heating.

3.5 Color

Whiteness of surimi gels was affected by egg white protein addition and applied heating conditions (Fig. 5). In the absence of egg white protein, whiteness of GF gels was lowest with a pinkish appearance with lower L* and higher a* values (data not shown). LF exhibited the highest whiteness, which is its distinct feature. Addition of egg white proteins only reduced whiteness of LF gel, but not surimi from other species.

Ohmic heating improved whiteness of LF gels when compared to water bath heating. Lightness value mainly depends on protein network. The rearrangement of water-protein interactions under ohmic heating regimes could be different from that induced by water bath heating. A more translucent appearance was observed in ohmically-heated gels. Moreover, non-enzymatic browning reactions seemed to be limited during ohmic heating. The lower b* value (data not shown) tended to result in surimi gel cooked with shorter heating times. Since surimi contained 60 g kg^{-1} sugar was heated for 30 min using water bath, this would have more chance to allow browning reaction between sugar and protein than those heated for 1 and 3 min using ohmic heating. It was noted that the rapid heating method with shorter heating time could preserve color as compared to water bath heating.

4. Conclusions

Ohmic heating significantly improved gelation of tropical surimi as it thermally inactivated endogenous proteinases in a rapid fashion. An improvement in texture is more pronounced in proteinase-laden surimi of GF and LF. Ohmic heating providing a higher rapid heating of 16.7 V cm^{-1} is not favorable for TB surimi, which is known to contain less proteolytic enzymes among tropical surimi. Addition of egg white proteins may not be necessary when cooked rapidly using ohmic heating. Disulfide bond formation played an important role in enhancing textural properties of tropical surimi gels through ohmic heating. Ohmic heating also improved water holding capacity and whiteness of gels. Based on our

findings, conventional surimi gel preparation using water bath was found to underestimate the value of all tropical surimi. For accurate assessment of the gel value, surimi should be cooked rapidly using ohmic cooker when it is to be used for crabstick production.

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535

List of Table

Table 1 TCA-soluble oligopeptide of threadfin beam (TB), bigeye snapper (BS), goatfish (GF) and lizardfish (LF) pastes and gels without (No-EW) and with 10 g kg^{-1} egg white proteins.

List of Figure

Fig. 1 SDS-PAGE pattern of lizardfish, LF (a) and goatfish, GF (b) gels prepared from various treatments. P; surimi paste incubated at 65 ^{o}C for 1 h, No-EW; without egg white proteins, +10 g kg^{-1}EW; with 10 g kg^{-1} egg white proteins, WB; water bath heating, OH 6.7 OH 16.7; ohmic heating with applied voltage gradient of 6.7 and 16.7 V cm^{-1}, respectively.

Fig. 2 Breaking force (a) and deformation (b) of threadfin beam (TB, ▥), bigeye snapper (BS, ■), goatfish (GF, ▩) and lizardfish (LF, ◧) gel prepared from various treatments. No-EW; without egg white proteins, +10 g kg^{-1}EW; with 10 g kg^{-1} egg white proteins, WB; water bath heating, OH 6.7 OH 16.7; ohmic heating with applied voltage gradient of 6.7 and 16.7 V cm^{-1}, respectively. Bars represent the standard deviation from 10 determinations. Different letters (a-n) on the bars indicate a significant difference at $P<0.05$.

Fig. 3 Total SH concentration of surimi pastes (P) and threadfin beam (TB, ▥), big eye snapper (BS, ■), goatfish (GF, ▩) and lizardfish (LF, ◧) gels prepared from various treatments. No-EW; without egg white proteins, +10 g kg^{-1}EW; with 10 g kg^{-1} egg white proteins, WB; water bath heating, OH 6.7 OH 16.7; ohmic heating with applied voltage gradient of 6.7 and 16.7 V cm^{-1}, respectively. Bars represent the standard deviation from 4 determinations. Different letters (a-p) on the bars indicate a significant difference at $P<0.05$.

561 Fig. 4 Water holding capacity of threadfin beam (TB,), bigeye snapper (BS,),
562 goatfish (GF,) and lizardfish (LF,) gel prepared from various treatments.
563 No-EW; without egg white proteins, +10 g kg^{-1}EW; with 10 g kg^{-1} egg white
564 proteins, WB; water bath heating, OH 6.7 OH 16.7; ohmic heating with applied
565 voltage gradient of 6.7 and 16.7 V cm^{-1}, respectively. Bars represent the
566 standard deviation from 4 determinations. Different letters (a-m) on the bars
567 indicate a significant difference at $P<0.05$.
568 Fig. 5 Whiteness of threadfin beam (TB,), bigeye snapper (BS,), goatfish (GF,)
569 and lizardfish (LF,) gels prepared from various treatments. No-EW; without
570 egg white proteins, +10 g kg^{-1}EW; with 10 g kg^{-1} egg white proteins, WB;
571 water bath heating, OH 6.7 OH 16.7; ohmic heating with applied voltage
572 gradient of 6.7 and 16.7 V cm^{-1}, respectively. Bars represent the standard
573 deviation from 5 determinations. Different letters (a-i) on the bars indicate a
574 significant difference at $P<0.05$.
575